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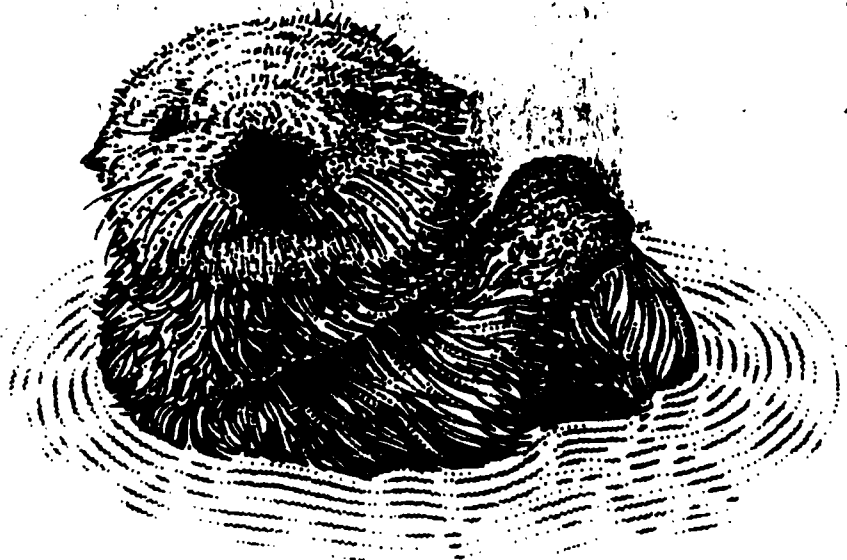
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PROCEEDINGS


Renal Function, A Possible Indicator of Stress in Dolphins

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Stress is a poorly defined condition which is experienced by all mammals. The term usually has been used with negative connotations which gives the impression that all stress is bad. Every animal, including man, undergoes some form of stress daily. Subjection to stress at frequent intervals stimulates organic mechanisms to react in ways essential to survival. How well the animal reacts to stress dictates how well it can adjust to changes of its environment. In this sense stress is useful. Individual organ systems, especially the endocrine system, respond to neurohumoral signals in diverse ways. The net physiological effect is a response to stressors that maintains homeostasis near normal.

Renal mechanisms play a major role in maintenance of the internal environment at a constant composition and volume. Changes in urine solute and water reflect demands on the kidney to maintain plasma normalcy. If changes in renal function occur during stress, understanding the physiological basis of these changes would enhance health management decisions.

Research methods for our experiments were as follows. Five bottlenose dolphins (*T. truncatus*) were used in six simulated transport studies. Two of the dolphins were female and three were male animals. On the day of an experiment food was withheld. At 0800 the animal was lifted from its pen, placed on a padded stretcher and control blood samples collected. Each animal served as its own control. The padded stretcher was placed in an animal transporter and moved to a laboratory which had controlled environment. The animal was catheterized for urine and blood samples. Self adhering electrodes were attached to the skin to record the electrocardiogram (ECG) and monitor heart rate. Respiration was monitored visually. When all instrumentation was completed blood and urine samples were taken, the ECG, heart rate and respiratory rate recorded. This was zero time. Samples were collected hourly for 6 or 12 hours then the dolphin was returned to its pen. The next day at 0800 (24 hours) and 1400 (30 hours)

blood samples were taken again. Blood, serum or plasma were assayed for cortisol, aldosterone, plasma renin activity (PRA), prostaglandins, osmolality, electrolyte concentrations, and pH (whole blood). A complete blood count (CBC) and a panel 20 which included creatinine and blood urea (BUN) were also performed. Urine volume was measured to obtain minute volume in order to calculate the glomerular filtration rate (GFR) using the creatinine clearance technique. Laboratory analysis of urine included sodium (Na^+), and potassium (K^+) concentrations as well as osmolality. A conventional urinalysis (UA) was also performed.

Results were as follows: serum cortisol concentration averaged 55 nmol/L in control (C) samples. By zero (0) time which varied from 1½ to 2 hours (the time for transport and instrumentation of the dolphin) the cortisol had risen to 80 nmol/L. From that point there was a steady increase until the 9th hour, at which time it was 125 nmol/L. There was then a decline to about 85 nmol/L at the 12th hour. At 24 hours the concentration was 53 nmol/L (same as the control) and at 30 hours 28 nmol/L.

Plasma aldosterone followed a pattern similar to cortisol. Control levels were 2 ng/dl. By 0 time the concentration had risen to 12 ng/dl and at the 12th hours was 16 ng/dl. By 24 hours it had declined to 3 ng/dl and at 30 hours to 2 ng/dl.

Plasma renin activity (PRA) changed much slower over time when compared to the adrenal steroids. Control values were 1 ng/ml/hr. At 0 time the concentration was 1.5 ng/ml/hr. There was nearly a straight line increase from that point reaching 5 ng/ml/hr at 12 hours. By 24 hours it was 1.5 ng/ml/hr and at 30 hours 1.2 ng/ml/hr.

The animals conserved water during the simulated transport which was reflected in the concentration of urine. Osmolality was 1200 mOsm/kg H_2O at 0 time

and rose in a linear fashion to 1800 mOsm/kg H₂O by 12 hours. Thus, the capability for dolphins to reabsorb salt free water was clearly demonstrated.

Another parameter of importance was the ratio of excreted sodium to potassium, expressed as $[Na^+]/[K^+]$. From experiments in dogs and cats, we have found the normal ratio to be between 1 and 2. In an animal which has been stressed (hypovolemia, dehydrated) with normal kidney function the ratio can decrease to as low as 0.2. A high ratio (greater than 8) suggests primary renal failure or inadequate aldosterone secretion (Addison's disease). The ratio in our studies decrease rapidly with time at first, then became steady. The control value of 2.4 fell to 0.21 by the 3rd hour and remained near that value until the 12th hour. The decrease with time was almost inverse to the rise in aldosterone and indeed reflected the effect of the latter hormone. Sodium was conserved at the expense of potassium secretion. This was an appropriate response because sodium is essential to preservation of plasma volume. When comparing the urinary $[Na^+]/[K^+]$ to the fractional excretion of sodium (the amount of filtered sodium actually excreted in urine) we found the curves were nearly identical. However, the urine sodium to potassium ratio was far easier to obtain since it required only the electrolyte concentrations of a small volume (1 ml) of urine.

Discussion

What appeared to be happening was a reversal of the immersion phenomenon during which an animal (person) has a strong urge to urinate soon after being submerged in water. The basis for this urge is physiological. The pressure of the water compresses limbs and cavities which increases venous return. Receptors in the heart perceive this as hypervolemia and reflexly inhibit antidiuretic hormone (ADH) release. More solute-free is excreted and urine volume increases.

The dolphin is normally in an aqueous environment and usually at neutral buoyancy. As soon as we raised it out of the water, pressure on its body is reduced. Although the animal was well supported in a comfortable padded stretcher it was no longer at neutral buoyancy. The receptors of the dolphin perceive the new environment as one in which it was hypovolemic. Even though there was no fall in measured blood pressure the physiologic responses were appropriate for the situation. Cortisol and aldosterone rose and subsequently produced a more concentrated urine as sodium and solute free water were conserved. This tended to maintain blood volume. The low urine $[Na^+]/[K^+]$ was appropriate for this response.

In conclusion, the dolphin is capable of existence out of water for extended periods as demonstrated by responses to 6 and 12 hour simulated transports. This was achieved by invoking appropriate physiological endocrine and renal mechanisms which maintain homeostasis. When returned to water all the dolphins swam normally and ate as soon as they were offered fish. Measurements of important parameters which reflect stress had returned to normal by 24 and 30 hours.

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